

CALCIUM ARSENATE DUSTING AS A CAUSE OF APHID INFESTATION

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INTRODUCTION

The cotton louse or cotton aphid (*Aphis gossypii* Glover), like the "green bug" (*Toxoptera graminum* Rond.) and others of its relatives, becomes exceptionally abundant and destructive in certain years when climatic and other factors favor its multiplication. In the South the louse was sporadic in its occurrence in 1922, more widely distributed in 1923, and prevalent in 1924, 1925, and 1926.

Since 1922 the Delta laboratory of the Bureau of Entomology, at Tallulah, La., has conducted, among other projects, extensive investigations concerning the biology and control of this aphid on cotton,¹ some of the results of which afford a solution to a problem that has been a puzzle to entomologists.

In recent years it has been noticed, particularly by entomologists actively engaged in the suppression of the boll weevil, that excessive applications of calcium arsenate to cotton fields may be followed by a conspicuous and often injurious increase of the cotton louse. Accordingly it becomes a matter of importance to learn whether the poison, while acting directly to kill one pest, indirectly favors the multiplication of another. It was necessary, therefore, to make a thorough experimental study of the subject from the economic point of view to determine the conditions under which the cotton louse multiplies to an injurious degree and the most effective means of controlling it along with the boll weevil.

¹ The investigations have yielded voluminous results, of which those that form the subject of this circular have been tested to a degree that warrants their publication. The field operations in the vicinity of Tallulah, La., in 1924, 1925, and 1926 were conducted by the junior writer. Supplementary entomological studies in 1925 and 1926 were made by the senior writer, assisted by F. A. Glick and temporary assistants.

METHODS EMPLOYED

The field investigations of this complex problem necessitated each year the use of 178 plats, totaling about 65 acres. These field operations are not reported in detail, but are summarized in this circular. The experiments were conducted in triplicate on three fields representing three types of soil. The experimental cuts were laid out where plant growth was uniform. The seasonal history of each plat was recorded in the form of plant counts, measurements of plant heights, counts of squares, blooms, and bolls, records of weevil and of aphid infestation, weights of cotton yields, and daily meteorological records.

The applications of calcium-arsenate dust ranged from 3 to 7 pounds or more per acre, according to the stage of growth of the plants, with an average dosage of 6 pounds. For the control of the aphids nicotine mixtures of various strengths were used. The weaker concentrations of nicotine (1.58 to 2.36 per cent) gave poor control unless heavy applications (11.69 pounds per acre) were made. One application of a stronger concentration (2.62 per cent), used at the rate of about 11 pounds per acre, sufficed, however, to eliminate the heaviest infestation of aphids.

The percentages of aphid infestation were computed by the following method: (1) The percentage of plants infested was determined by an examination of 200 successive plants located at each of three points; one near the middle of the field and one near each end of it. (2) The percentage of infestation for each of the 600 plants examined was estimated by comparison with a standard series of photographs representing 10 grades of infestation, 1 to 10 per cent, 11 to 20 per cent, and so on. (3) Then the percentage of infested plants was multiplied by the average percentage of infestation per plant, as in the following example: Percentage of infested plants, 50; average infestation per plant, 50 per cent; total infestation, 50 per cent of 50 per cent, or 25 per cent.

AVERAGE EFFECTS OF EXCESSIVE APPLICATIONS OF CALCIUM ARSENATE

In order to determine whether excessive applications of calcium arsenate would give rise to an excessive aphid infestation, 4-plat experiments were made on each of the three fields. Plat 1, the check, received no treatment. Plat 2 was poisoned with calcium arsenate from June 18 to August 13, inclusive, at intervals of four or five days, the total number of applications being 14. This treatment was intended to control the boll weevil. Plat 3 received the same calcium-arsenate treatment as plat 2; in addition it received from two to five applications of nicotine dust, the number of applications depending upon the character of the particular mixture used, and the dates of application being determined by the severity of the aphid infestations which they were intended to reduce. Plat 4 received, as its only treatment on each cut, the same kind of nicotine treatments as plat 3. Thus the effects of the nicotine alone could be ascertained.

An excessive number of applications of calcium arsenate on plat 2 raised the degree of aphid infestation steadily, beginning June 26,

to a peak of 97.1 per cent on July 29. Thereafter, under continued applications, the infestation remained between 89 and 100 per cent. Meanwhile the infestation of the check plat was only 12.8 per cent on July 7, and thereafter remained below that figure until toward the end of the season. Nicotine dust, used whenever the infestation was increasing badly on the arsenate-treated plats (No. 3), served at once to check or to reduce greatly the infestation. The same kind of nicotine treatments, without calcium arsenate, on plat 4 served to hold the degree of infestation below that of the check throughout the season.

AVERAGE EFFECTS OF APPLICATIONS OF CALCIUM ARSENATE REPEATED THROUGHOUT THE SEASON

In order to determine the effects of calcium arsenate applied repeatedly throughout the season, the poison was applied to cotton plants from May 20 to August 29, inclusive, at intervals of four days, the total number of applications being 25. This experiment was similar in scope to the one just described, except that the applications were continued over a longer period.

On the plants treated with calcium arsenate the aphid infestation began to rise on June 28, increased uniformly to a peak of 100 per cent on July 18 and 19 and August 1 (the experiments being in triplicate), and maintained an average of 98 per cent or more until about August 8, thereafter falling until the end of the season. During the height of the infestation on the check plats, however, the highest point reached was only 10.3 per cent.

These experiments demonstrated that repeated applications of calcium arsenate brought an immense increase in the aphid population. They showed also that the earliest applications caused no material increase in infestation. It became necessary, therefore, to determine the extent to which the increase depended upon the time of application and the relation between the increase and the number of applications. These questions were answered by means of other experiments.

SEASONAL EFFECTS OF CALCIUM ARSENATE ON APHID INFESTATION

In order to ascertain exactly at what time during the season applications of calcium arsenate dust would cause an increase in aphid infestation, experiments were made on three cuts, each consisting of 46 plats. The first and the last plat (at opposite ends of each cut) were used as checks, and therefore received no treatment. The remaining plats were poisoned from two to eight times at intervals of usually four, and never more than five, days, beginning on various dates from May 20 to August 9. These were designed to show the effects at any period of the season of any number (from two to eight) of successive applications of the dust. On each of the three cuts each experiment was made on six different plats.

Two applications of calcium arsenate made on May 20 and 24, respectively, produced no effect. Two applications, June 2 and 6, were likewise ineffective. Two applications, June 14 and 18, increased the aphid infestation, which began to rise abruptly on June 30 and attained a peak of 63.7 per cent on July 18. Meanwhile the

infestation on the check plats was only 10.3 per cent. Later applications (two per plat) on other plats were followed by somewhat lower percentages of increased infestation.

TABLE 1.—*Aphid infestation on cotton following two successive applications of calcium arsenate, Tallulah, La., 1924*

Plat No.	Date of application	Maximum infestation		Interval between first dusting and maximum infestation
		Date attained	Percentage	
4.....	May 20, 24.....	(1).....		Days
11.....	June 2, 6.....	(1).....		
18.....	June 14, 18.....	July 18.....	63.7	34
25.....	July 1, 5.....	do.....	51	17
32.....	July 14, 18.....	Aug. 1.....	57.3	18
39.....	July 30, Aug. 4.....	Aug. 15.....	47	16

¹ No increase.

The relation of the effects of calcium-arsenate applications to the time of the season appeared most clearly in the 2-application series.

Applications early in the season (up to June 14) were not followed by an increase in the aphid population, while treatments from June 14 to July 9 were followed by the greatest increases during the entire season. Applications made during the last three weeks of July operated to induce moderately high infestations, about 40 to 70 per cent as high as those following applications late in June.

The general increase in the infestations which followed applications made during August was greater than that following applications late in July, but less than that which followed the applications beginning June 14. Applications on any dates between June 14 and July 14—regardless of their number (two to eight)—brought the peak of infestation on July 18. Applications on July 14 and 18, or on July 14, 18, and 22, however, produced the maximum effect later; that is, on August 1. In fact, after July 14, the later the applications were made, the later the maximum infestations appeared.

The interval between the time of insecticide application and maximum aphid infestation was by no means constant, as shown in part in the last column of Table 1. For example, the two applications in June produced their maximum effect in about one month, whereas later applications were followed by maximum aphid infestations in a little more than two weeks.

EFFECT OF THE NUMBER OF APPLICATIONS OF CALCIUM ARSENATE

The same experiments gave data also on the degree of aphid infestation in relation to the number of applications of calcium arsenate.

From two to six treatments, when applied before June 14, were ineffective in reducing aphid infestation.

From one to seven treatments, when applied from June 14 to July 9, were followed by increase of infestation, the maximum infestations ranging from 28 to 95 per cent and rising with the number of applications.

Eight treatments, ending July 14, were followed by an 80 per cent infestation.

Applications, from two to eight in number, in July and August did not bring an increase of infestation that was proportionate to the number of applications, though a tendency toward such an increase was manifest.

It was found that two applications induced the greatest increase in aphid population when they were made between June 14 and June 18; three treatments resulted in the maximum increase when applied June 14 to 23; four when applied June 14 to 27; five when applied June 14 to July 1; six when applied June 14 to July 5; seven when applied June 14 to July 9; eight when applied June 14 to July 13.

EFFECTS OF THE USUAL APPLICATIONS OF CALCIUM ARSENATE

It was necessary to determine whether calcium arsenate, when applied according to the regular, or commercial, program for weevil control, would give rise to an injurious aphid infestation; and if so, to determine whether or not a single application of nicotine mixture, made at the proper time, would control the louse. Experiments with these objects in view were conducted in triplicate on 4-plat cuts.

Plat 1 on each cut was left as a check. Plat 2 received calcium arsenate only. Three applications were made at intervals of about four days, beginning when 10 per cent of the squares were punctured; two more treatments followed about two weeks later, as further weevil infestation made them necessary. On plat 3, in addition to calcium-arsenate treatments like those on plat 2, a single application of nicotine mixture was made when the aphid infestation became excessive. Plat 4 received the same nicotine treatment as plat 3, but no calcium arsenate.

Following the first three dustings of calcium arsenate, the aphid infestation on plats treated with calcium arsenate alone began to rise rapidly, and the last two applications brought it up to 100 per cent (August 20), where it stayed. On plat 3, however, one application (August 23) of nicotine mixture (lime base with 2.62 per cent nicotine, about 10 pounds per acre) wiped out the infestation. The same kind of nicotine treatment also destroyed the aphids on plat 4. Meanwhile the check plat showed almost no infestation. In fact the infestation on that plat did not exceed 4 per cent during the entire period of insecticidal treatment.

NATURAL CONTROL OF THE COTTON LOUSE

The experiments that have been summarized in the foregoing sections demonstrated that excessive applications of calcium arsenate on cotton plants act to increase the aphid population. In explanation of this result the most plausible hypothesis is that the poison diminishes one or more of the factors that normally check the multiplication of the aphids. These adverse factors are many and involved, and their action and interaction constitute a complex subject for study.

The principal external influences that affect the multiplication of plant lice are (1) the condition of the host plant, (2) climatic factors, (3) disease, and (4) enemies.

CONDITION OF THE HOST PLANT

Some of the influences that affect the nutrition of a host plant affect indirectly the reproduction of its aphid inhabitants. These factors need not, however, be considered in connection with the present subject, for nutritional conditions in the experimental cotton fields were as uniform as it was possible to have them.

CLIMATIC FACTORS

Temperature and moisture influence the reproduction of plant lice (1) indirectly, as by affecting the food plants, or by influencing the development of disease or insect enemies; and (2) directly, as through evaporation, or the effects of winds or rains. During the experiments the climatic influences did not affect the results essentially. It was observed, however, as had already been demonstrated in breeding experiments, that high temperatures with low humidity were apparently favorable to the multiplication of the aphids.

DISEASE

The most inimical influence to which aphids in general are exposed is the widespread and contagious disease due to the fungus known as (*Empusa*) *Entomophthora aphidis* Hoff., which, when epidemic, sweeps off myriads of aphids. The disease may appear after a rain at any time during the growing season, and is most prevalent under the combined influences of moisture and warmth. In the dry summer of 1924 the disease was absent from the experimental plats. In 1925 and 1926 it was present at times, but affected plant lice indiscriminately—without regard to applications of calcium arsenate.

MISCELLANEOUS ENEMIES

As checks upon aphid increase, certain miscellaneous enemies, as birds, spiders, and mites, are relatively unimportant in comparison with the host of insect enemies.

INSECT ENEMIES OF THE COTTON LOUSE

In order to get information on the numbers of such insects as might influence the abundance of the cotton louse, collections were made systematically of insects occurring on the cotton plants. The collections were made in each plat by "sweeping" all the plants of three rows (each 250 feet in length) with a net 18 inches in diameter. Naturally, the manner in which the sweeping was performed (as regards, for example, the number of strokes per row and the length and duration of each stroke) was made as nearly uniform as possible. The sweepings, made every four days throughout the season, yielded an immense quantity of material. This method of collecting, admittedly inexact for quantitative studies, was nevertheless sufficiently accurate to show whether a given species of insect was increasing or diminishing in numbers from day to day.

A great number of insects are concerned with the cotton louse in one way or another. Some 60 species are known to influence the reproduction of the pea aphid (*Macrosiphum*) *Illinoia pisi* Kalt., either as predators or as primary or secondary parasites. No doubt the cotton louse is affected by an equally large number of species. However, for present purposes, only the dominant enemies of the cotton louse need be considered.

COCCINELLIDAE

The lady beetles, both as larvae and adults, are the most efficient predacious enemies of plant lice in general. Their numbers on a plant or in a field are, in a rough way, a measure of the numbers of plant lice present. They are capable of checking the increase of the aphids to a great extent, though their importance in this respect is likely to be overestimated; for the lady beetles, unaided by other natural agencies of control, are unable actually to exterminate the aphids. The beetles do not become common until aphids are abundant, and their prolificacy is far behind that of the plant lice. Thus, numerous as the beetles may be, they always lag behind their prey on account of unequal reproductive ability. Another reason is that Coccinellidae are by no means limited to a diet of plant lice but feed also on various other insects or arthropods, and often consume large quantities of pollen grains and fungus spores.

The most abundant coccinellid in the experimental plats was *Hippodamia convergens* Guér. On account of its feeding habits, abundance, and wide distribution this is generally the most important beneficial lady beetle in the United States. Next in point of numbers came the well-known *Ceratomegilla fuscilabris* Muls. (formerly *Megilla maculata*). *Coccinella novemnotata* Hbst. was very common. *Cycloneda munda* Say (*Coccinella sanguinea*) occurred frequently. Four species of *Scymnus* (*S. americanus* Muls., *S. loewii* Muls., *S. brullei* Muls., and *S. fraternus* Lec.) were taken on cotton; the larvae of these small lady beetles were abundant and important enemies of the cotton louse.

CHRYSIDAE

The predacious larvae of lacewing flies are everywhere recognized as important checks upon the multiplication of plant lice. The species taken in the collections, both as adults and as larvae, were *Chrysopa oculata* Say and *C. rufilabris* Burm.

SYRPHIDAE

Syrphid flies and their larvae were omnipresent in the cotton fields, the commonest species being those of aphidophagous habits. Of these the most abundant species collected was *Mesograpta polita* Say, which, though known to feed on pollen, particularly that of corn, feeds also on plant lice. Next in point of abundance came *Mesogramma gurgus* Walk.

HYMENOPTEROUS PARASITES

By far the most efficient insect enemy of the cotton louse was found to be the parasite *Lysiphlebus testaceipes* Cress. It appeared with the lice in the spring and was present in maximum numbers during the latter half of June and again during the first two weeks of August.

Another important primary parasite of the cotton louse was *Pachyneuron siphonophorae* Ashm. This was most abundant during the first week of July, the last two weeks of August, and in September and October; these were times when *L. testaceipes* was uncommon.

EFFECTS OF CALCIUM ARSENATE ON PREDATORS

It might be thought that calcium-arsenate dusting possibly favored the multiplication of the cotton louse by poisoning its chief predatory enemies, larvae and adults of Coccinellidae and larvae of Syrphidae and of Chrysopidae. To an observer they were not conspicuously fewer in number following applications of calcium arsenate. Systematic sweepings gave definite information on the subject, particularly sweepings made on plats treated with calcium arsenate and on check plats just before dusting and afterwards at intervals of 30 minutes, 5 or 6 hours, 24 hours, etc.

When the results of the sweepings were tabulated and compared with the curves showing the degree of aphid infestation it became evident that all these predators increased in numbers as the infestation increased. This was conspicuously so in the case of *Hippodamia convergens* Guér, the most abundant predator.

For the purpose of recovering any insects that might possibly be killed by calcium arsenate, cloths were spread under a number of cotton plants and held down under each plant by a fencelike frame of boards 4 feet square. Immediately before dusting, these "trays" were free from insects; after dusting, they were examined every few hours and for several days. The result was that from 10 of these trays only 12 adults of *Hippodamia convergens*, 5 syrphid larvae, and 3 Scymnus beetles were collected during two months. A few ants and spiders entered the trays accidentally; a few insects fell from plants when the latter were disturbed; and that was all. The predators were not killed by the dusting.

EFFECTS OF CALCIUM ARSENATE ON PARASITES

During the course of biological studies of *Lysiphlebus testaceipes* Cress., the effect of calcium-arsenate dusting on this most important insect enemy of the cotton louse was accidentally discovered. Lice parasitized by this species were scraped from cotton leaves, incidentally with more or less calcium arsenate, and kept in Petri dishes for the study of emergence. The parasites, upon emerging, struggled about in the dust, became coated with it, and died in from 10 to 30 minutes. They died also in the open air in the presence of calcium arsenate. Subsequent experiments showed that the parasites were similarly killed by other dusts, some of which were inert. The results of these tests are presented in Table 2.

TABLE 2.—Results of experiments with dusts fatal to parasites of the cotton louse at Tallulah, La., 1925

Dust used	Lysiphlebus testaceipes		Pachyneuron siphonophorae	
	Parasites	Average time needed to cause death	Parasites	Average time needed to cause death
	Number	Minutes	Number	Minutes
Calcium arsenate.....	39	21.3	75	64
Calcium hydroxide.....	12	33.7	10	110
Calcium carbonate plus 2 per cent Paris green.....	38	38.1	-----	-----
Calcium carbonate.....	39	72.0	66	119
Cornstarch.....	44	77.5	30	540

Under a microscope the calcium-arsenate dust could be seen on the legs and body (abundantly when there was dew on the leaves), and the parasites often drew their feet through their mouths. Grains of calcium arsenate were not observed in the spiracles, and most of the grains were too large to enter them, but the spiracles were sometimes more or less covered by the grains. The calcium hydroxide was, of course, caustic. The calcium carbonate with 2 per cent of Paris green (used for dusting *Anopheles* larvae) killed *Lysiphlebus* much more rapidly than did calcium carbonate alone. Cornstarch, an inert substance, acted most slowly, gradually clogging the spiracles and the mouth parts. In the cotton fields, immediately following dusting with hand guns or airplane, adults of *Lysiphlebus* became rather scarce for 8 or 10 hours; they were undoubtedly blown about by the blasts of dust, and perhaps in many instances they were killed or incapacitated.

In May, before the cotton plants were dusted, the cotton louse was often heavily parasitized by *Lysiphlebus*, but after dusting began there was a noticeable decrease in parasitism.

Pachyneuron siphonophorae Ashm. was an important check on the increase of the cotton louse at times during the season when *Lysiphlebus testaceipes* was scarce. *Pachyneuron* was killed in the same manner as *Lysiphlebus* by calcium arsenate on the cotton leaves. It was likewise killed by dusts of other kinds, as calcium hydroxide, calcium carbonate, cornstarch, and flour. The reactions of these parasites to those substances were the same as those of *Lysiphlebus*. The average periods required to cause death are given in Table 2. It will be noted that these dusts operated less rapidly on *Pachyneuron* than on *Lysiphlebus*.

One effect, therefore, of dusting with calcium arsenate was to favor the multiplication of the cotton louse by reducing the numbers of its most important parasitic enemies.

PHOTOTROPISM OF THE COTTON LOUSE

Before parasites appeared, however, the cotton louse was much more abundant on dusted plants than on undusted. Winged lice appeared on the former a few hours after dusting, but not on the latter;

and two dustings, three days apart, induced a moderate infestation in a week or so. This had to be accounted for.

A clue was obtained when the senior writer, while following the men who were dusting the experimental cotton rows with hand guns, saw a winged female louse fly like a shot at a patch of calcium arsenate on a leaf, and remain there. The same thing was observed several times afterwards and led to experiments on the phototropism of the louse, which are here summarized.

In an uninfested part of a cotton field a single plant was dusted with calcium arsenate. Migrant aphids soon arrived, and in 8 or 10 days, with two dustings, a moderate infestation was under way, though the surrounding undusted plants remained uninfested. This experiment was repeated several times with the same result. A heavy infestation never developed, as predators of the cotton louse concentrated upon these isolated infested plants and rapidly destroyed the lice.

The same experiment was made by dusting segregated cotton plants with either calcium carbonate, cornstarch, or flour. These white dusts likewise induced infestation by aphids. The flour (applied as usual when dew was on the leaves) dried and caked on the leaves, causing the plant to wilt, but not before winged aphids had reached the plant.

Experiments in dusting were then made on a large scale for the comparison of calcium arsenate with another dust that should be white but inert. Cuts of cotton were laid out, each of about 3 acres, divided into three equal plats: (1) A check plat, (2) a plat dusted with calcium carbonate, and (3) a plat dusted with calcium arsenate. In 1925 the dusted plats developed eventually 100 per cent infestation, possibly for the reason that the only immediately available carbonate contained 2 per cent of Paris green (for use in dusting swamp areas by airplane for the destruction of larvae of malaria mosquitoes).

In 1926 pure calcium carbonate was used on the No. 2 plats of the two cuts—M and K. Plats 2 and 3 in cut M were dusted seven times from July 3 to August 14, an average of 17 pounds of calcium carbonate and 6 pounds of calcium arsenate being used each time. Plat 2, however, had to be poisoned with Paris green for the leaf worm on August 11. Plats 2 and 3 of cut K received no Paris green and were dusted eight times, from June 11 to August 14, each time with 13 pounds of carbonate and 8 pounds of arsenate, respectively. The percentages of initial and final infestation are shown in Table 3.

TABLE 3.—*Aphid infestation following dusting with calcium carbonate and calcium arsenate in large-scale experiments, Tallulah, La., 1926*

Cut	Plat No.	Dust used	Original infestation		Final infestation, Aug. 20
			June 11	July 3	
			Per cent	Per cent	Per cent
M	1.....	None (check).....		0.7	38.0
	2.....	Calcium carbonate.....		.3	78.0
	3.....	Calcium arsenate.....		.1	95.0
K	1.....	None (check).....	3.3		18.0
	2.....	Calcium carbonate.....	2.0		25.2
	3.....	Calcium arsenate.....	1.3		69.0

The high infestation in plat 2 of cut M was possibly due to the Paris green.

The plats treated with calcium arsenate and calcium carbonate always became infested at the same time after dusting, and the infestation developed equally on the two plats until parasitism began; thereafter infestation became gradually greater on the arsenate plat than on the carbonate plat.

As another phototropic test, plants were dusted with calcium arsenate that had been colored green. These plants remained uninfested for a time, whereas adjacent plants dusted with white arsenate became infested in three days. In from 5 to 10 days, however, the green arsenate faded to almost white in the sunlight and infestation set in.

It has often been observed that aphid infestation becomes more intense on small areas of dusted cotton (as on experimental plats) than on large dusted areas of hundreds or thousands of acres. An explanation of this fact would be that the positive phototropism of the aphids causes them to concentrate on white areas of cotton that are relatively small, whereas in areas in which all the cotton in a district is whitened the aphids have no opportunity for selection.

SUMMARY

Excessive applications of calcium arsenate are often followed by heavy infestations of the cotton louse. In the investigations to determine the cause, a number of experiments were made on the effects of frequent and prolonged treatments of plats of cotton under varying seasonal conditions.

The agencies of natural control are described, and the relation between the insect enemies of the aphid and the dusting is pointed out. In experiments it was found that a heavy aphid infestation is built up by the killing of hymenopterous parasites when they emerge in the presence of the arsenical. They were killed also, though more slowly, by calcium hydroxide, calcium carbonate, or cornstarch. Predatory insects, especially coccinellids, which destroy great numbers of aphids, were not affected by the calcium arsenate.

The initial infestations were found to be due to the positive phototropic reaction of winged females to the white deposit of calcium arsenate. Other white dusts, such as calcium carbonate, starch, or flour, were also found to attract them.

ORGANIZATION OF THE UNITED STATES DEPARTMENT OF AGRICULTURE

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